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14. ABSTRACT

This report describes the synthetic approaches toward a potent microtubule stabilizing natural product, elutherobin, utilizing tandem Diels-Alder reaction/Grob-type fragmentation reaction as key steps. During the course of these studies, large scale preparation of *bis*-diene and successful activation of the secondary alcohol were achieved. Due to the difficulty in Sm-mediated fragmentation and capricious nature of the tandem Diels-Alder reaction, our original route did not prove amenable to an efficient synthesis of eleutherobin. A revised route using a -elimination pathway was subsequently investigated and functionalization by Wharton rearrangement reaction was also studied.

15. SUBJECT TERMS
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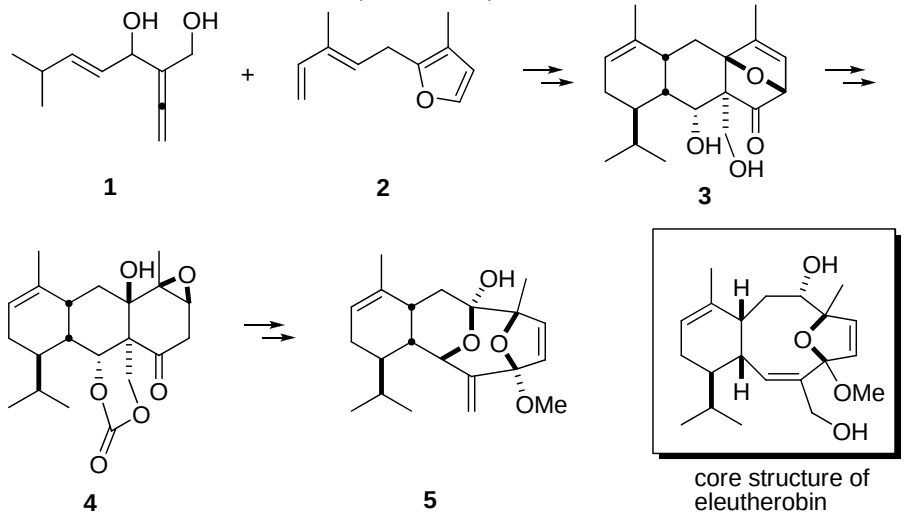
1. Introduction

Limited availability and highly potent anticancer activity of eleutherobin inspired the synthetic communities. With the aim of establishing a stable synthetic route to eleutherobin and development of prostate-cancer specific analogues, we studied the synthetic methods to the eleutherobin core using tandem Diels-Alder reaction. Our synthetic approaches as well as two novel chemical reactions found in the synthetic efforts are described.

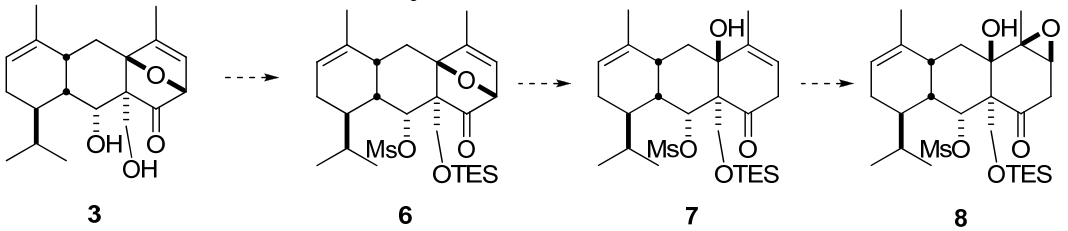
2. Body

Our previous approach[1)] demonstrated that a tandem Diels-Alder reaction of bis-dienophile precursor **1** and bis-diene **2** followed by Grob fragmentation of carbonate **4** gave tetracycle **5** which could be utilized in the formation of carbon skeleton found in eleutherobin (Scheme 1).



Scheme 1. Previous approach

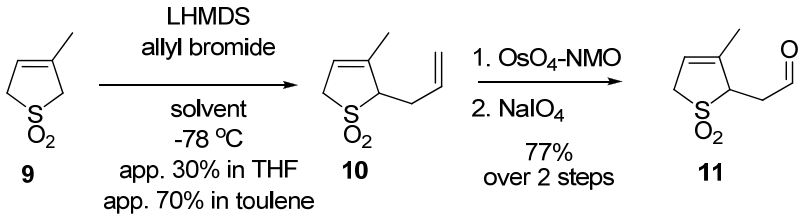
However, the key fragmentation reaction of **4** gave only the undesired product due to the presence of another activated primary hydroxyl group. It was then clear that selective activation of the secondary alcohol must be achieved for the desired cleavage to occur. Accordingly, we turned our attention to installing the activating group on the secondary hydroxyl group before ring opening of the diol **3** (Scheme 2). The resulting mesylate **6** could be easily converted into tertiary alcohol **7** based upon our previous successful C-O cleavage reactions. Regioselective epoxidation would then produce tetracycle **8** that satisfied all the requirements for the selective fragmentation in eleutherobin core synthesis.



Scheme 2

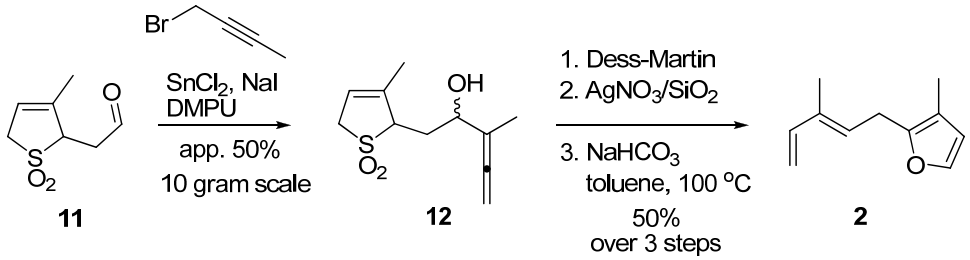
To test our revised synthetic design, we originally followed our previously reported synthetic procedure. Soon we noticed a number of problems posed in large scale preparation of synthetic intermediates, especially the *bis*-diene **2**, which would be required to complete the total synthesis. One of the biggest challenges in our previous synthesis of the *bis*-diene **2** was the preparation of the known sulfolene aldehyde **11**[2)] on large scale (Scheme 3). Conventional conditions required extremely low temperature (-100^{o}C, THF) to prevent

polymerization and polyalkylation of the 3-methyl sulfolene **9**. However, maintenance of such a low temperature in large scale reactions (typically, more than 10 grams) was practically unmanageable due to the temperature fluctuation during base addition and the desired product was usually obtained in poor yield. To address this issue, we tested various conditions. Changing the solvent from THF to toluene showed a dramatic increase in the reaction yield. Under the optimized condition (toluene, -78 ^{o}C), we were able to obtain the allylation product **10** on multigram scale in 70% yield reproducibly. However, the sulfolene aldehyde **11** was unstable as we found a substantial amount of intractable black tar after storage under Argon (1 week, 4^{o}C). Thus, it was critical to use the aldehyde **11** in the next reaction as soon as possible.



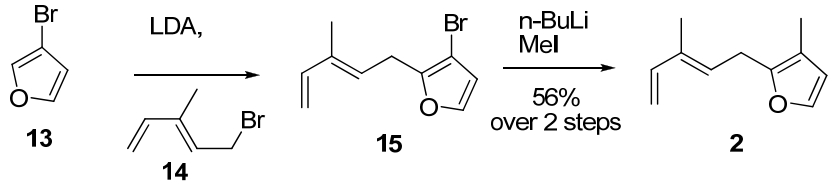
Scheme 3

With a successful solution to the large-scale preparation of the sulfolene aldehyde **11**, we set to the synthesis of *bis*-diene unit **2** on large scale (Scheme 4). In the large-scale preparation of allenic alcohol **12**, complete removal of DMPU by washing with aqueous NH_4F solution was not successful presumably due to repetitive Et_2O extraction to ensure the complete transfer of the desired product into the organic layer. Substantial amounts of DMPU remained even after column chromatography due to its high volume in the crude mixture. However, we found little detrimental effect of the residual DMPU in the next Dess-Martin oxidation reaction when we performed a test reaction using the crude mixture. We then chose to proceed with the crude allenic alcohol **12** for the next reaction but the unstable nature of allenic ketone obtained in the Dess-Martin oxidation also required prompt cyclization using silver nitrate. Thus, the combination of three steps (addition, oxidation, and cyclization) without column chromatography facilitated our large scale preparation with minimization of decomposition. Thermal extrusion of sulfur dioxide proceeded without any difficulty and we were able to produce the *bis*-diene **2** in up to 1 gram quantities using the aforementioned modified procedure. However, it must be noted that the *bis*-diene **2** was unstable and polymerization occurred extensively on storage for more than 1 week.



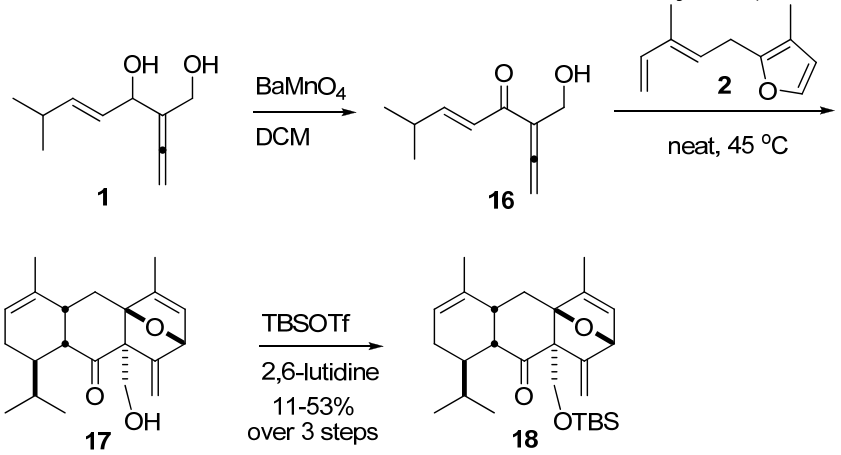
Scheme 4

It became apparent that this 7-step sequence to prepare the unstable *bis*-diene **2** was not a suitable route and we needed to find a better method of preparation. We envisioned that Schlosser's double alkylation procedure to prepare rosedfuran could be applied to preparation of our *bis*-diene **2**. C_2 alkylation of bromofuran **13** with the known bromodiene **14**[3)] produced alkyl bromofuran **15** and subsequent C_3 methylation using the same protocol furnished the desired *bis*-diene **2** in multigram quantities (Scheme 5). No further attempt to optimize the reaction conditions was made since we were able to obtain enough amount of the *bis*-diene **47** (app. 2 grams) from a single experiment.



Scheme 5

With large quantities of the *bis*-diene **2** and the *bis*-dienophile precursor **1** in hand, we set to study our tandem Diels-Alder reaction. However, the reported yield of this key reaction was not easily reproducible in our hand and we usually obtained the desired Diels-Alder adduct **18** in 11-53% yield (Scheme 6).



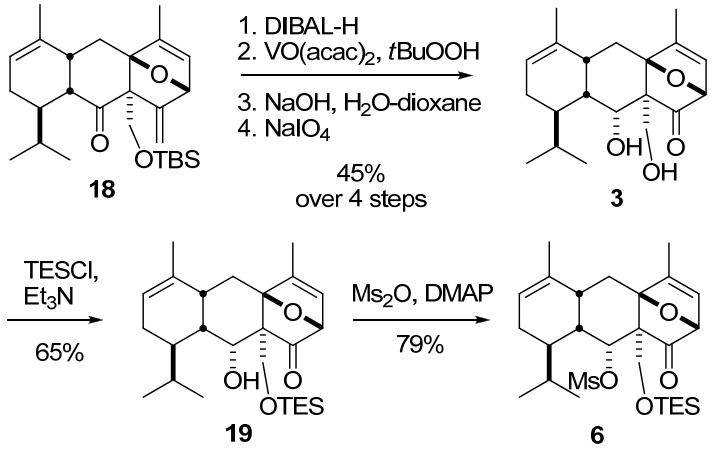
Scheme 6

The major problematic step was the selective oxidation of allenic diol **1** to allenic ketone **16** using $BaMnO_4$. Regardless of the origin of the reagent, the oxidation product was always accompanied by the formation of significant amounts of other unidentifiable compounds and conversion of the diol **1** to the allenic ketone **16** was very slow. Excess amounts of reagent had to be added and progressive decomposition of the resulting allenic ketone **16** was always observed. Other oxidants (MnO_2 or $BaMnO_4$-$CuSO_4$-alumina) or different solvents (Et_2O, ethyl acetate) did not improve the conversion rate. Excess amounts of $BaMnO_4$ also made the isolation of the crude allenic ketone **16** very difficult. The reason for the capricious nature of the oxidation reaction is still not clear to us since each experiment gave inconsistent results (Table 2.1). In most runs, the amount of *bis*-diene **2** employed was less than half the molar equivalent of the *bis*-dienophile precursor **1** due to the slow conversion to allenic ketone **16** and its decomposition. A good (53%) yield was obtained in a single run (2 g scale) but this was not reproducible on 6 g scale. To our disappointment, the largest-scale reaction gave none of the allenic ketone **16** but only decomposition was observed.

diol **1**	$BaMnO_4$	*Bis*-Diene **2**	**18** (yield)
0.5g	7 eq	0.4 eq	31%
0.78g	12 eq	0.45 eq	25%
0.8g	10 eq	0.56 eq	11%
1g	15 eq	0.3 eq	12%
2g	8 eq	0.4eq	53%
6g	6 eq	N/A	N/A

Table 2.1

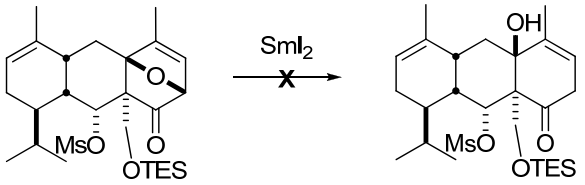
Despite the capricious yields in the tandem Diels-Alder reaction, we were able to obtain enough material to test our idea on selective activation. Transformation of Diels-Alder adduct **18** into the diol **3** went smoothly according to our previous protocol even on large scale (Scheme 7). We then decided to protect the primary hydroxyl group as the corresponding triethylsilyl ether due to its smaller size than the original TBS group. The formation of the silyl ether went smoothly to give the monoprotected alcohol **19**. After much experimentation, mesylate **6** was produced from diol **3** in 51% overall yield using excess Ms_2O and DMAP with mild heating. Thus, only C-O cleavage reaction was the only remaining task to prepare Grob-type fragmentation substrate.



18 3 19 6

Scheme 7

Unfortunately, reductive cleavage of ketone **81** using SmI_2 as a single electron donor failed to produce tertiary alcohol **82** under a variety of reaction conditions (Scheme 2.25).



6 7

Scheme 8

Change of temperature without additives did not alter the result of the ethereal bond cleavage reaction (Table 2.2). Treatment with additives like HMPA or MeOH did not produce the desired tertiary alcohol and only starting material was recovered. When catalytic amounts of NiI_2 were added, starting material disappeared but an unidentifiable complex mixture was obtained. Finally, when the proton donor dimethylethanolamine was used to suppress the Lewis acidity of Sm(III), we were able to isolate very small quantities of a new compound whose structure appeared to be the carbonyl reduction product, based on ^{1}H NMR analysis and transformation of the compound to the original ketone **6** by Dess-Martin oxidation.

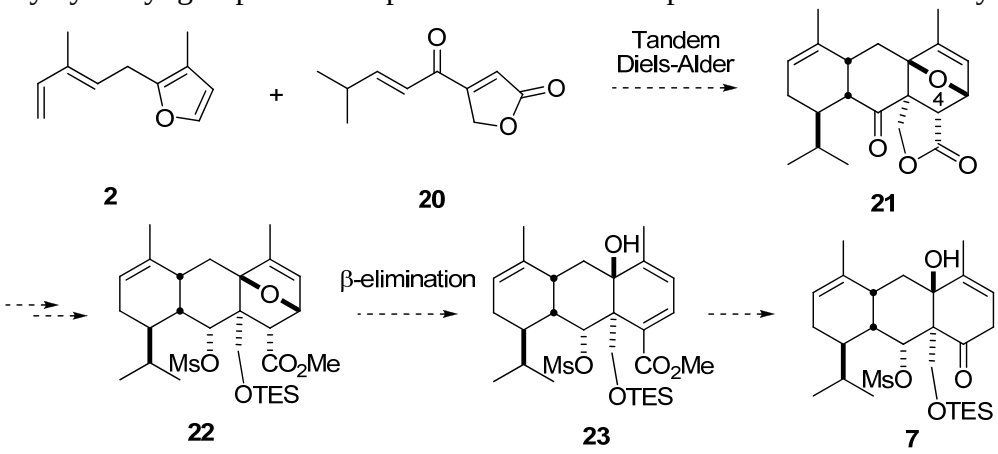
Additive	Temperature	Result
None	-78 ^{o}C	SM recovered
None	25 ^{o}C	SM recovered
None	50 ^{o}C	SM recovered
HMPA (3eq)	25 ^{o}C	SM recovered
MeOH(3eq)	25 ^{o}C	SM recovered
NiI_2 (cat.)	-20 to 25 ^{o}C	Complex mixture
$(CH_3)_2NCH_2CH_2OH$ (3eq)	-20 to 25 ^{o}C	Complex mixture

Table 2.2

We still do not have a clear explanation for these disappointing results. Subtle differences in substrate structures could be responsible for the observed failure. Contrasting results of ring opening reaction in a similar oxanorboranone system are precedented in the work of Cossy.[4]

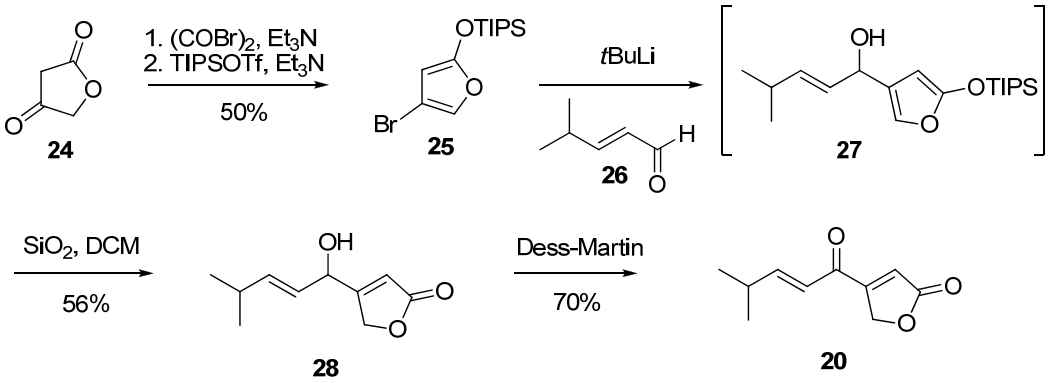
The failure of the C-O cleavage reaction led to the development of another idea that tertiary alcohol **7** could be derived from ester containing compound **23** by hydrolysis/Curtius rearrangement (Scheme 9). The

conjugated ester **23** then could be obtained from oxanorboranyl ester **22** by C-O bond cleavage reaction through β-elimination. Consequently, we revised the design of our tandem Diels-Alder reaction using a different *bis*-dienophile **20.** In this revised approach, *bis*-dienophile **20** would possess a doubly activated olefin as the more reactive dienophile and the C_4 carbonyl group present in the desired Diels-Alder product **21** could promote the epoxide opening by β-elimination. The stereochemical outcome of tandem Diels-Alder reaction product **21** could be expected based upon our previous results in the synthetic studies on eleutherobin. Selective activation of the secondary hydroxyl group would be possible based on the previous successful mesylation of **19.**



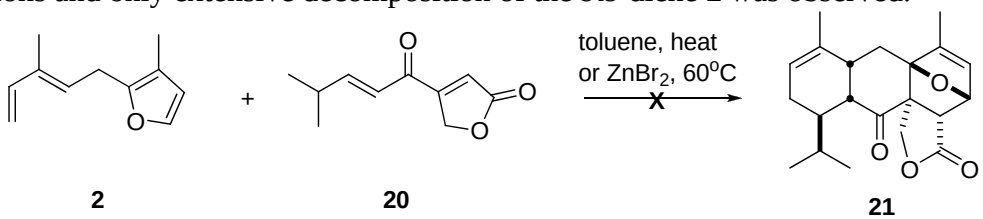
Scheme 9

We then set out to synthesize the *bis*-dienophile **20** (Scheme 2.29). 2-silyoxy-4-bromfuran **25** was prepared from tetronic acid **24** by vinyl bromide formation using oxalyl bromide followed by treatment of TIPSOTf. Metal-halogen exchange using *t*BuLi and addition of the lithiated furan to 4-methyl pentenal **26** provided the intermediate 4-alkyl-2-silyloxyfuran **27**. Deprotection of the silyl group could not be achieved by conventional TBAF or pTSA treatment. However, the desired γ-hydroxy lactone **28** was obtained by treatment of crude **27** with silica gel in DCM in 56% yield. Dess-Martin oxidation of the secondary alcohol **28** gave the *bis*-dienophile **20**.



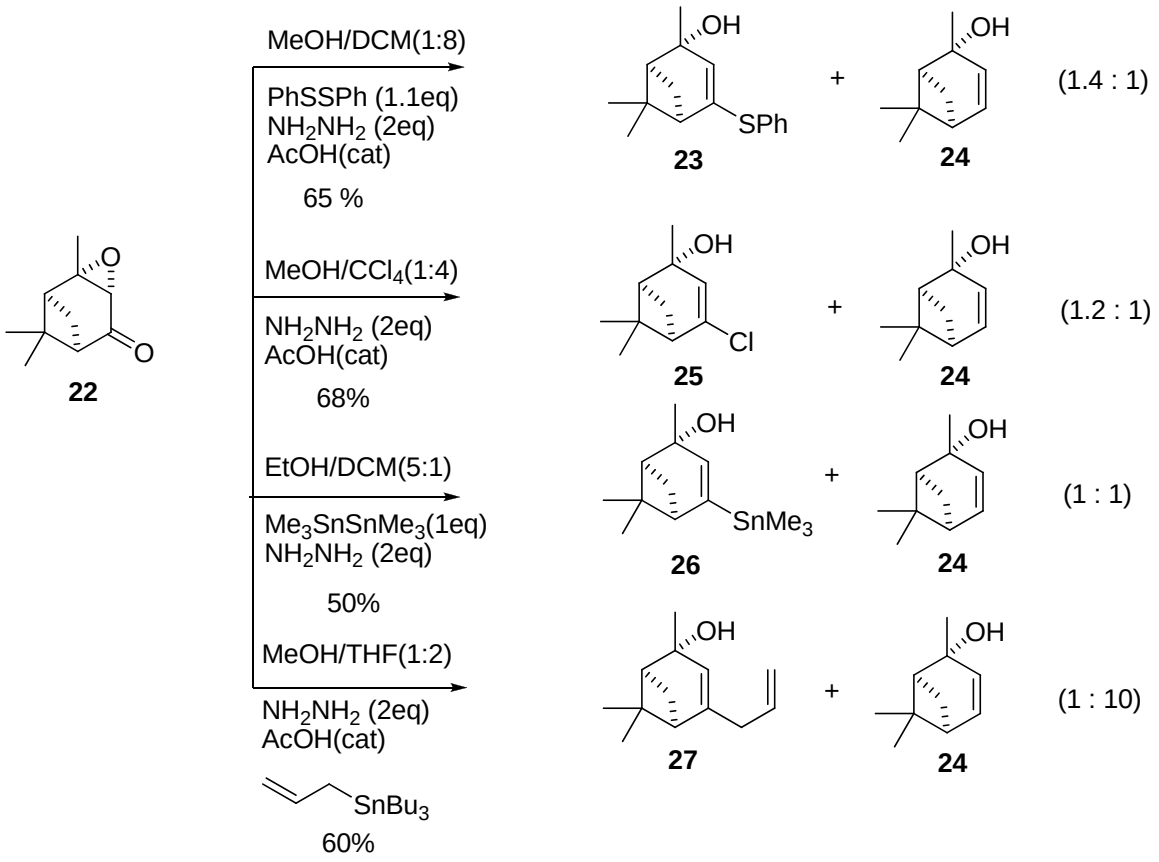
Scheme 10

With this material in hand, we attempted our tandem Diels-Alder reaction with the same *bis*-diene **2** (Scheme 11). Unfortunately, no Diels-Alder adduct **21** was obtained under thermal or Lewis acid ($ZnBr_2$) mediated conditions and only extensive decomposition of the *bis*-diene **2** was observed.



Scheme 11

Failure of the tandem Diels-Alder reaction in the β-elimination route led us to further investigate a-heteroatom cleavage reaction of ketone – Wharton rearrangement.[5] In our study, simple application of the Wharton rearrangement would only lead to the formation of diene alcohol that is not suitable for our purpose to retain the original carbonyl functionality. We then envisioned trapping of the intermediate vinyl radical[6] with a suitable reagent could produce functionalized allylic alcohol. We tested this idea on a model system – verbenone oxide **22** and the expected functionalized allylic alcohol (**23**, **25**, **26**, and **27**) were easily obtained.



Scheme 3. Wharton functionalization of epoxy ketone **10**

One of the issues in this transformation was the formation of normal Wharton rearrangement product **24**. The formation of **24** was suppressed when monosilylated hydrazine[7] was used but the overall reaction yield was poor. This novel Wharton functionalization is quite versatile as it provides a direct method to prepare vinyl functionalized allylic alcohols in a facile manner.

3. Key Research Accomplishment

- Large scale synthesis of *bis*-diene
- Successful preparation of advance intermediate for Gron fragmentation
- Discovery of Wharton functionalization

4. Reportable outcomes

Poster presentation
"Synthetic studies on the Taxol-like cancer chemotherapeutic agent"
Innovative Minds in Prostate Cancer Today (IMPaCT) meeting, Atlanta, GA, Sep 2007

Ph. D. degree will be awarded to the PI (2008 December)

5. Conclusion

During the three years of this grant, we have developed a procedure suitable for the large scale synthesis of *bis*-diene. Successful activation of the secondary alcohol was achieved but the next C-O cleavage reaction failed. During the study on the C-O cleavage reaction, a novel chemical transformation based upon Wharton rearrangement was discovered. This reaction would lead to a number of synthetically useful intermediates and give further insight on the traditional Wharton reaction. Even though the successful formation of the core structure in eleutherobin failed, the synthetic studies conducted during the grant period were extremely valuable in development of other synthetically novel and efficient routes to elecutherobin.

"So What?"

A novel Wharton functionalization was discovered and it could be a very powerful tool in the synthetic community if combined with organometal catalyzed C-C bond forming reactions.

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7. Appendices

N/A